# Oahuhawaiiana kazukolinda gen. et sp. nov. (Turbellaria, Tricladida, Maricola), a New Freshwater Planarian from Honolulu, Oahu Island, Hawaii, U.S.A.

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**ABSTRACT** — A new species of the new genus *Oahuhawaiiana* (Turbellaria, Tricladida, Maricola) from the Manoa Stream, Honolulu, Oahu Island, Hawaii, is described: *Oahuhawaiiana kazukolinda* Kawakatsu et Mitchell, gen. et sp. nov. This pigmented species with two eyes inhabiting an epigean freshwater locality seems to be a marine relict. It has been placed tentatively in the Maricola family Bdellouridae Diesing, 1862. The spherical cocoon without a stalk of this species contains 4 juveniles.

### **INTRODUCTION**

Mr. Ichirô Tanaka, as a special trainee at the East-West Center in Honolulu, Hawaii, from June, 1966, to July, 1967, collected a number of specimens of a small planarian and its cocoons from Manoa Stream, which is located within the city (lat. 21°19'N and long. 157°50'W; alt. approximately 300 m). Most of the samples were fixed with Bouin's fluid and sent to Kawakatsu for identification, together with brief sketches of the general appearance of some live specimens and cocoons. Mr. Tanaka also cultured several live specimens and cocoons in his laboratory (water temp. 22-25°C, pH 6.2-6.4) [1]. Tanaka's collection was designated as Kawakatsu's Specimen Lot No. 596. Because of the scarcity of wellpreserved, fully sexually mature specimens in this material, Kawakatsu waited long to obtain additional specimens from this locality.

Although numerous specimens of *Dugesia* species were collected from the same and two additional localities in Honolulu by Mr. Tanaka

(in 1966 and 1967) and Dr. Y. Hirao (in 1976 and 1977), the first species in question was never recollected [1, 2]. Recently a *Dugesia* species from Honolulu was identified as *D. dorotocephala* (Woodworth, 1897) [3, 4]. At the time the *Dugesia* species was studied, Kawakatsu made a very careful reexamination of his serial sections of the first species and succeeded in a general interpretation of the genital anatomy and reconstruction of the copulatory apparatus. After discussions, we have concluded that this animal is a peculiar retrobursal form of triclad turbellarian representing a new genus and species. We tentatively assign this monotypic genus to the family Bdellouridae.

#### MATERIALS AND METHODS

Two preserved specimens and cocoons were photographed in Kawakatsu's laboratory. Twenty-two specimens were cut in serial sagittal sections  $(7-8 \ \mu m)$  and were stained with Delafield's hematoxylin and erythrosin. A single sexually mature specimen was prepared in whole mount without staining.

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# SPECIES DESCRIPTION

# Genus Oahuhawaiiana Kawakatsu et Mitchell gen. nov.

Order Tricladida Suborder Maricola or Retrobursalia Family B d e l l o u r i d a e Diesing, 1862 [5] *Definition* Bdellouridae of normal body form; without adhesive disc; inhabitant of freshwater; head of low triangular form with a pair of very slight auricular expansions; pigmented; with two eyes and a single pharynx; testes rather few,



FIG. 1. Oahuhawaiiana kazukolinda gen. et sp. nov. from the Manoa Stream, Honolulu, Hawaii. A-C: Photographs of 3 preserved specimens, dorsal views (Specimen Lot No. 596). D: Cocoons. E: Preserved whole-mount specimen (No. 596 w). F: Photomicrograph of the sagittal pharyngeal section (No. 596 h; holotype). 1 and 3, longitudinal fibers of outer muscle zone; 2, circular fibers of outer muscle zone; 4, longitudinal fibers of inner muscle zone; 5, circular fibers of inner muscle zone. phl, pharynx lumen.

extending posteriorly to the level of a pair of female genital pores; penis normal, consisting of bulb and papilla; common sperm duct entering bulbar cavity; single ejaculatory duct normal; a pair of ovaries in the usual anterior position; common genital pore opening into the common genital antrum; a pair of copulatory bursae situated posterior to the common genital pore, each having a narrow, ventral, bursal canal opening separately into the posterior part of the female genital canal; female genital canal opening into the common genital antrum anteriorly and into the single seminal bursa posteriorly; seminal bursa connected by a narrow seminal bursaintestinal duct to an intestinal cross-link: ovovitelline ducts separately entering the middle part of the female genital canal.

*Type species* The type and only known species is named and described as follows:

# Oahuhawaiiana kazukolinda Kawakatsu et Mitchell sp. nov.

The specific name we have chosen for this unusual planarian has been compounded from the given names of Mrs. Kawakatsu (Kazuko) and Mrs. Mitchell (Linda), whose help to us through the years has been invaluable during the course of our studies.

**Description** This is a small, slender, pigmented species with two eyes. Live, sexually mature specimens measure 6 to 8 mm in length and 0.5 to 0.8 mm in width. Preserved animals are somewhat contracted (Fig. 1A–C, E). The head is of a low triangular form with a bluntly pointed anterior end. The auricles are represented only as a pair of gentle expansions occurring posterolaterally on the head. There is no "neck" or narrowing of the body behind the head. The posterior end of the body is bluntly pointed (Fig. 1A–C, E).

The ground color of the dorsal surface of the animal is light yellowish brown. The midline is striped with a rather wide, longitudinal yellowish band. The body margin and the areas above the pharynx and copulatory apparatus are of a lighter hue (Fig. 1A-C, E). Numerous microscopic, brown pigment spots are scattered on the

dorsal surface (Fig. 1E). The ventral side is light yellow.

The dorsal epithelium is much thicker than the ventral. The frontal adhesive glands are well developed (Fig. 3A).

The general position and arrangement of organs are shown schematically in Figure 2.

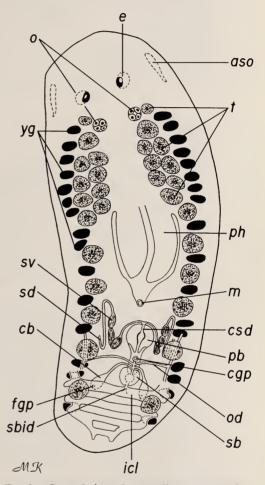


FIG. 2. General view of a sexually mature specimen of Oahuhawaiiana kazukolinda gen. et sp. nov., a composite schematic drawing from the whole mount (No. 596 w) and several serial sagittal sections (No. 596 a, h, p, and r). aso. auricular sense organ; cb, copulatory bursa; cgp, common genital pore; csd, common sperm duct; e, eye; fgp, female genital m, mouth icl, intestinal cross-link; pore; pb m od, ovovitelline duct; o, ovary; bulb; ph, pharynx; sb, seminal sbid, seminal bursa-intestinal duct; miducal vesicle; t, testis; yg, yo!' i and

Two rather large eyes are situated on the dorsal about, or slightly less than, one-third the width side of the head. The distance between them is of the head at the level of the eyes. Each eye

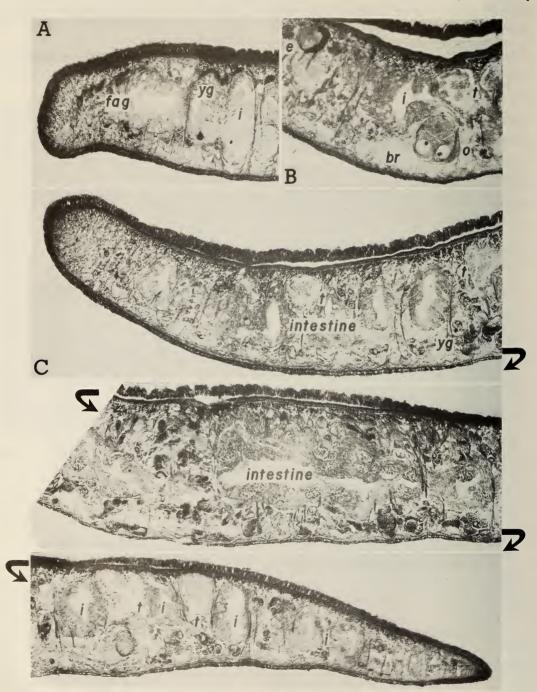


FIG. 3. Oahuhawaiiana kazukolinda gen. et sp. nov., sagittal section (No. 596 h; holotype). A: Head. B: Anterior part. C: Near midsagittal section. br, brain; e, eye; fag, frontal adhesive gland; i, intestine; o, ovary; t, testis; yg, yolk gland.

is enclosed in an elliptical, pigment-free ocular area (Figs. 1A-C, E, 2). A nonpigmented auricular sense organ is conspicuous on each posterolateral side of the head; each is of the shape of a willow leaf.

The pharynx is situated behind the middle of the body and measures nearly one-fifth of the body length. The common genital pore is situated in the midventral line at the level of the middle of the postpharyngeal region (or slightly anterior to this level) (Figs. 1E, 2). A pair of female genital pores (bursal pores or vaginal pores) open on each side of the midventral line posterior to the level of the common genital pore (Fig. 2). However, these pores cannot be perceived in whole, preserved specimens even when examined with the higher magnifications of a binocular stereomicroscope.

The anterior intestinal trunk bears 8 to 10 pairs of lateral branches. Each posterior trunk has 10 to 12 short branches. In the caudal region, 2 or 3 pairs of these branches are extended medially and unite to form transverse intestinal connections, or cross-links (Fig. 2). The inner musculature of the pharynx consists of a moderately thick layer of circular fibers adjacent to the ciliated epithelium of the pharyngeal lumen and a thin layer of longitudinal ones. The outer musculature of the pharynx consists of an outer, thin layer of longitudinal fibers; an inner, thin layer of circular ones; and a third, loose and inconspicuous layer of longitudinal fibers (Fig. 1F).

A pair of moderately large ovaries occur in the ventral space between the third or fourth lateral branches of the anterior intestinal trunk. Each ovary contains 3 to 5 ova (Figs. 2, 3B). Yolk glands (or vitellaria) are distributed throughout the body in the surrounding parenchyma (Figs. 2, 3A and C).

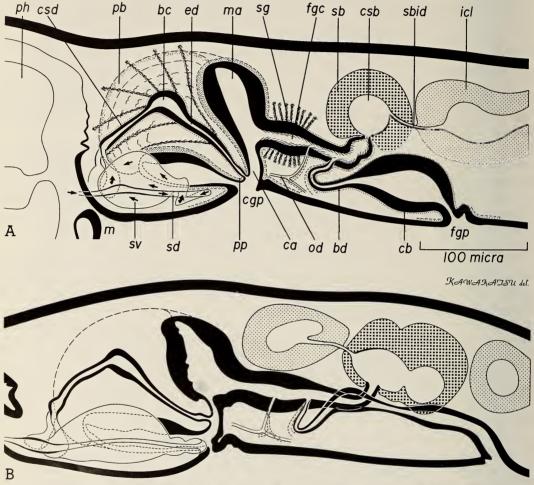
The testes are rather few, of moderate size, and located dorsally. In the prepharyngeal region, they are arranged on either side of the midline in two longitudinal rows. In the pharyngeal and postpharyngeal regions, they are arranged almost in a single row on either side of the body. The testes extend from a level just anterior to the ovaries to near the posterior level of the female genital pores (Figs. 2, 3B and C, 7A–D). Their total number is estimated to be about 25 to 30 (of which about 20 are located in the prepharyngeal region).

Of 22 specimens sectioned and examined, only 4 proved to be fully sexually mature (Lot No. 596 a, h, p, and r). Nine of the other specimens were also in sexual development but not fully mature. Although both the penis and common genital pore were present in these latter animals, the female genital pores and bursae were not differentiated (Lot No. 596 i-m, o, q, s, and u), although parts of the bursal canal were evident in some. The remaining 9 specimens had no sexual organs.

Figure 4 (A and B) shows sagittal views of the copulatory apparatus of 2 specimens (Lot No. 596 h, the holotype; and p). Photomicrographs of the parts of the copulatory apparatus of 9 specimens (including non-fully mature animals) are also shown in Figures 5 (A–J), 6 (A–J) and 7 (A–D).

The penis consists of a very large, hemiglobose bulb and a large, moderately long, conical papilla of symmetrical shape (Figs. 4A and B, 5A-H, 6A-J). Both the bulb and papilla are weakly muscular. The two sperm ducts run posteriorly to about the anterior level of the common genital pore, where they abruptly reverse their curve anteriorly. At about the level of the mouth they course inwardly to form remarkably developed spermiducal vesicles on each side of the body between levels just posterior to the mouth and just anterior to the middle of the penis papilla (Figs. 4A and B, 5I and J). On the anterolateral sides of the penis bulb, each spermiducal vesicle ascends and forms a narrow tube. These tubes converge in the posterior part of the bulb uniting to form a single, long, slightly widened duct (common sperm duct) that continues on into the bulb to open into a rounded, narrow lumen, or bulbar cavity (Figs. 4A and B, 6A and B, 7A-C, H). The bulbar cavity continues into the papilla as a narrow ejaculatory duct, which opens at the tip of the papilla (Figs. 4A and B, 5A-C, 6A-E, H-J). The penis lumen (both the bulbar cavity and ejaculatory duct) is lined with a glandular, missions epithelium. Below this epithelium, there and think layer of circular muscle fibers. The huldlogy

of the common sperm duct in the bulb is similar to that of other parts of the penis lumen. Moderately erythrophilous penis glands open into the penis lumen (Figs. 4A and B). The surface of the penis papilla is covered with a tall, glandular, nucleate epithelium, below which there are two layers of muscle fibers, one a slightly thickened layer of circular fibers and the other a



- FIG. 4. Oahuhawaiiana kazukolinda gen. et sp. nov., semidiagrammatic sagittal views of the copulatory apparatus. A: No. 596-h (holotype). B: No. 596-p (paratype). bc, bulbar cavity; bd, bursal duct; cb, copulatory bursa; ca, common genital antrum; cgp, common genital pore; csb, cavity of the seminal bursa; csd, common sperm duct; ed, ejaculatory duct; fgc, female genital canal; fgp, female genital pore; ich, intestinal cross-link; m, mouth; ma, male genital antrum; od, ovovitelline duct; pb, penis bulb; ph, pharynx; pp, penis papilla; sb, seminal bursa; sbid, seminal bursa-intestinal duct; sd, sperm duct; sg, shell gland; sv, spermiducal vesicle.
  - FIG. 5. Oahuhawaiiana kazukolinda gen. et sp. nov., copulatory apparatus (No. 596 h; holotype). A-H: Serial near midsagittal sections. I and J: Near midsagittal sections (No. 596 h). bc, bulbar cavity; bd, bursal duct; cgp, common genital pore; csd, common sperm duct; ed, ejaculatory duct; fgc, female genital canal; icl, intestinal cross-link; m, mouth; ma, male genital antrum; nc, nerve cord; od, ovovitelline duct; pb, penis bulb; ph, pharynx; pp, penis papilla; sb, seminal bursa; sg, shell gland; sv, spermiducal vesicle.

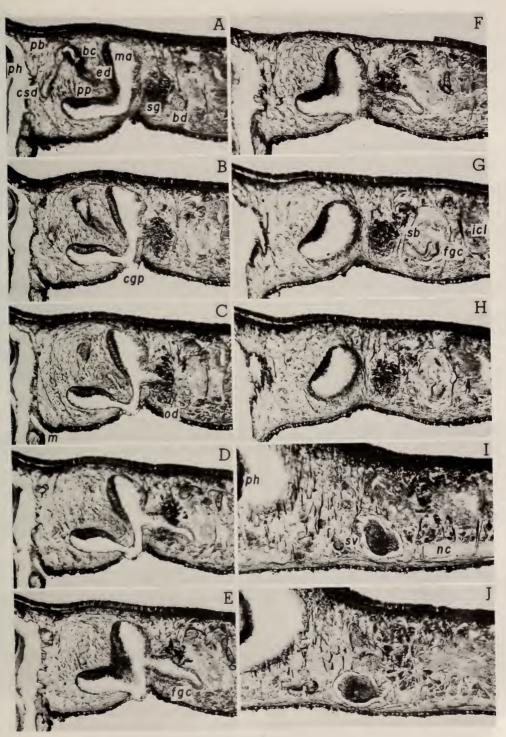


FIG. 5

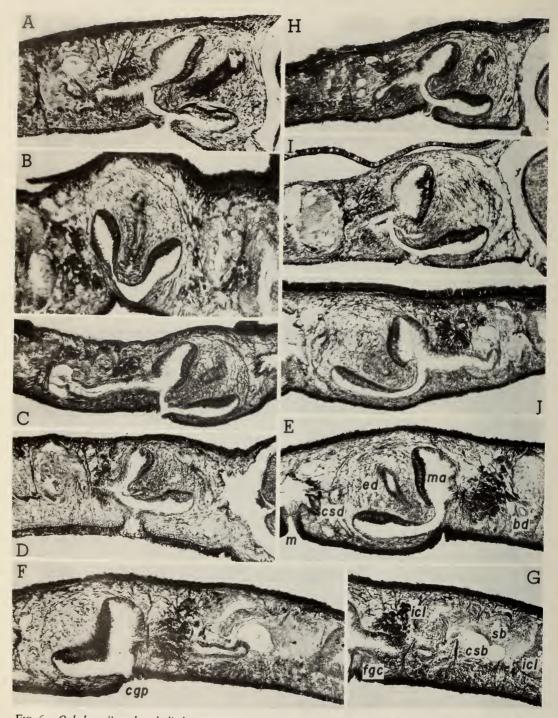


FIG. 6. Oahuhawaiiana kazukolinda gen. et sp. nov., copulatory apparatus of 8 paratypes. A: No. 596-a.
B: No. 596-i. C: No. 596-k. D: No. 596-m. E-G: No. 596-p. H: No. 596-q. I: No. 596-r.
J: No. 596-s. bd, bursal duct; cgp, common genital pore; csb, cavity of the seminal bursa; csd, common sperm duct; ed, ejaculatory duct; fgc, female genital canal; icl, intestinal cross-link; m, mouth; ma, male genital antrum; sb, seminal bursa.

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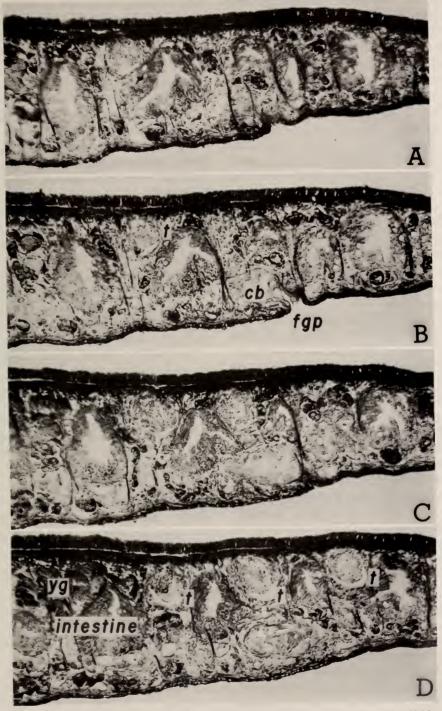


FIG. 7. Oahuhawaiiana kazukolinda gen. et sp. nov., copulatory apparatus (No. 596 h; holotype). A-D: Serial sagittal sections through the female genital pore. cb, copulatory bursa; fgp, female genital pore; t, testis; yg, yolk gland.

thin layer of longitudinal fibers. The male genital antrum is a wide, cup-shaped cavity that opens at the common genital pore posteroventrally. It receives the female genital canal via an opening in its midposterior region (therefore, the posteroventral part of the genital antrum seems to be a common genital antrum, which opens at a common genital pore) (Figs. 4A and B, 5A-H, 6A-J). The walls of the male and common antra are covered with a tall, glandular, nucleate epithelium, below which there are two muscle layers, a slightly thickened one of the circular fibers and an outer, thin one of longitudinal fibers. The glandular epithelium of the posterodorsal part of the male antrum is more developed than that in other antral regions.

The anatomy of the female part of the copulatory apparatus is complicated (Fig. 2). Each female genital pore leads into a moderately wide cavity, the copulatory bursa. It is an elongated spheroid (Figs. 4A, 7A–D), or it is club-shaped (Fig. 4B). Its inner wall is lined with a glandular, nucleate epithelium. The lining epithelium at the beginning of the bursa (near the female pore) is ciliated. Anteriorly, the bursae form long, narrow ducts (bursal ducts or bursal canals) that open separately into the posteroventral portion of the female genital canal (Figs. 2, 4A and B, 5A–D, H, 6E). Both the bursae and the bursal canals are surrounded by a thin coat of circular muscle fibers. Sperm masses occur in the bursal canals.

The female genital canal is a rather long and wide duct. It is lined with a ciliated, glandular, nucleate epithelium, below which there is a thin layer of circular muscle fibers. The female canal receives a pair of thin ovovitelline ducts midventrally (Figs. 4A and B, 5C). The wall of the anterior, slightly widened portion of the female canal is pierced by the ducts of heavily erythrophilic glands (shell glands) that open into the canal (Figs. 4A, 5A–H, 6A–J). This portion of the canal is filled with fine erythrophilic granules.

The posterior, terminal portion of the female genital canal opens into the widened cavity of a bursa-like organ, or seminal bursa. The seminal bursa is globose (Figs. 4A, 5G and H) or spheroidal (Figs. 4B, 6F and G) and is without a muscular coat. Its wall consists of tall, glandular, epithelial cells, of which most have insunk nuclei. The cavity of the seminal bursa is connected by a long, narrow duct (seminal bursa-intestinal duct) to an intestinal cross-link lying posterior to the seminal bursa (Fig. 4A) or to a cross-link lying to the anterior (Fig. 4B).

The egg-capsule, or cocoon, is spherical (0.3–0.4 mm in diameter) and does not bear a stalk (Fig. 1D).

Type series Holotype. One set of sagittal serial sections (Specimen Lot No. 596 h, 1 slide) will be deposited in the Department of Zoology, National Science Museum (Nat. Hist.), Tokyo. Two paratypes, a set of serial sagittal sections (No. 596 p, 1 slide) and 1 specimen as whole mount (No. 596 w), will also be deposited in the same Museum. The other slides, including several paratypes (No. 596 a, i-m, o, q-s, and u), and other slides of sexually immature specimens, as well as cocoons preserved in 70% ethanol, are retained by the senior author (Kawakatsu's laboratory, Fuji Women's College, Sapporo).

*Type locality* Manoa Stream (upper part of the Manoa-Palolo Drainage Canal), St. Louis Heights, Honolulu, Oahu Island, Hawaii, U.S.A. [4; see footnote].

Taxonomic remarks and differential diagnosis To the present, 8 species of retrobursal triclads inhabiting freshwater are known. They are as follows:

Balliania thetisae Gourbault, 1978, from Tahiti [6]; Debeauchampia anatolica Benazzi, 1981, from Asiatic Turkey [7]; Mitchellia sarawakana Kawakatsu et Chapman, 1983, from Sarawak in East Malaysia, Borneo Island [8]; Opisthobursa mexicana Benazzi, 1972 (=Dimarcus villalobosi Mitchell et Kawakatsu, 1972), and Opisthobursa josephinae Benazzi, 1976, from México [9-11]; Opisthobursa? sp. Kawakatsu et Mitchell, 1983, from Guatemala [12]; Rhodax evelinae Marcus, 1946, from Brazil [13]; and Eviella hynesae Ball, 1977, from Australia [14].

Of these, the genera *Balliania*, *Debeauchampia* and *Mitchellia* have been placed tentatively in the family Procerodidae Diesing, 1862 [5], suborder Maricola (=Retrobursalia). The genus *Opisthobursa* belongs to the Maricola family Dimarcusidae Mitchell et Kawakatsu, 1972 [10] (=Opisthobursidae Benazzi et Giannini, 1973 [15]) [12]. The genera *Rhodax* and *Eviella* have been placed tentatively in the family Dugesiidae Ball, 1974 [16], suborder Paludicola (=Probursalia). The 6 genera listed above are all monotypic, except *Opisthobursa*. A more detailed list of them including the type locality of each species and related publications will be found in a previous paper by Kawakatsu and Chapman [8].

Oahuhawaiiana kazukolinda described in the present paper is the ninth retrobursal form inhabiting a freshwater locality and is the first known freshwater species in the maricolan family Bdellouridae Diesing, 1862 [5]. To the present, 9 genera and 12 species are known in this family, including the species described herein. They are as follows:

1. Genus Bdelloula Leidy, 1851 [17]

1) Bdelloura candida (Girard, 1850) (olim Vortex candida) [18]. A commensal on the horseshoe crab Limulus polyphemus along the New England coast of the United States [see also 19–21].

2) Bdelloura propinqua Wheeler, 1894 [22]. A commensal on L. polyphemus [see also 20, 21].

3) Bdelloura wheeleri Wilhelmi, 1909 [20]. A commensal on L. polyphemus [see also 21].

2. Genus Syncoelidium Wheeler, 1894 [22]

4) Syncoelidium pellucidum Wheeler, 1894 [22].

A commensal on L. polyphemus [see also 20, 21].

3. Genus Synsiphonium Hallez, 1911 [23]

5) Synsiphonium liouvilli Hallez, 1911 [23]. Loc. Petermann Island in the Antarctic [23, 24] and Tierra del Fuego, South Argentina [25].

4. Genus Pentacoelum Westblad, 1935 [26]

6) *Pentacoelum fucoideum* Westblad, 1935 [26]. Loc. Brackish water in South Sweden.

7) *Pentacoelum caspium* Beklemischev, 1954 [27]. Loc. Caspian Sea (Kaspiiskoe More; brackish water), U.S.S.R.

5. Genus Nerpa Marcus, 1948 [28]

8) Nerpa evelinae Marcus, 1948 [28]. Loc. Beach in the vicinity of Santos, Brazil.

6. Genus Palombiella Westblad, 1951 [29]

9) Palombiella stephensoni (Palombi, 1938) (olim Synsiphonium stephensoni) [30]. Loc. Oudekraal and Port Nolloth, South Africa [30]; the Islands of Tristan da Cunha in the South Atlantic Ocean (ca. lat. 37°03'S and long. 12°18'W) [29].

7. Genus Siphoniella Westblad, 1952 [25]

10) Siphoniella anderssoni Westblad, 1952 [25]. Loc. Tierra del Fuego, South Argentina.

8. Genus Miava Marcus, 1954 [31]

11) *Miava evelinae* Marcus, 1954 [31]. Loc. South Pacific coast of South Chile.

# 9. Genus Oahuhawaiiana Kawakatsu et Mitchell, 1984

12) Oahuhawaiiana kazukolinda Mitchell et Kawakatsu, 1984. The new species in the new genus described herein. Loc. See foregoing section.

Of the 9 genera listed above 7 are monotypic. The genus Bdelloura includes 3 species. Of these (except for Oahuhawaiiana 12 species, 11 kazukolinda) are inhabitants of marine waters. if 2 Pentacoelum species from brackish water localities be included. The 3 Bdelloura species, all of them commensal on Limulus, are unpigmented, have the adhesive zone expanded into a caudal adhesive disk, and have numerous testes. Syncoelidium pellucidum, the other commensal on Limulus, is a pigmented form without adhesive organ and having a rather limited number of testes. The remaining 8 species are free-living. The species of the genera Pentacoelum (white in color and without eye-pigment), Nerpa (white in color) and Siphoniella, have only a pair of testes in the prepharyngeal region. Synsiphonium liouvilli has numerous testes. The species of the genera Palombiella (a pigmented form) and Miava (a pigmented form) have a rather limited number of testes.

A slight resemblance in the genital anatomy is found between *Miava evelinae* and *Oahuhawaiiana kazukolinda*. The former has an elongated, cornshaped penis with a lumen separated by diaphragm into a tubular bulbar cavity ejaculatory duct (the original author used the original Samenblase). Two posterior, spherical contactory bursa (Bursae seminales), each of which opens ventrally as a female genital pore (Vaginalporen), receive ovovitelline ducts (Ovovitellodukte) which form a short common duct opening into the female genital canal (weiblicher Genitalkanal) and into the seminal bursa (Uterus) [31, see figs. 53, 55 and 56 on p. 103]. These characters of the genital anatomy of *Miava evelinae* are, however, quite different from those of *Oahuhawaiiana kazukolinda*. More detailed descriptions of the general appearance, histology, and genital anatomy of the 8 genera (*Bdelloura, Syncoelidium, Synsiphonium, Pentacoelum, Nerpa, Palombiella, Siphoniella*, and *Miava*) are found in the revised key of the "Familie: Bdellouridae" [31, see pp. 35–36; see also 28].

Oahuhawaiiana kazukolinda, the present new species, is characterized as follows: living animal small (less than 8 mm in length), pigmented, with a pair of eyes; epigean freshwater inhabitant of the Manoa Stream in Honolulu, Oahu Island, Hawaii (known only from the type locality); head of low triangular form with a pair of gentle auricular expansions accompanied by a pigment-free sense organ on each; with a single pharynx; dorsal testes moderately large, 25 to 30 in total number, on either side extending from a level just anterior to the ovaries to the posterior level of the female genital pores; penis bulb large, hemiglobose in shape and weakly muscular with a narrow, rounded bulbar cavity into which a long common sperm duct enters (the two sperm ducts form welldeveloped spermiducal vesicles on each side of the bulb); symmetrical penis papilla large, moderately long, conical in shape, weakly muscular, and with a narrow ejaculatory duct; male genital antrum large, cup-shaped, and opening into the common genital pore; two ovaries in the usual anterior position (each ovary contains 3 to 5 ova); two female genital pores, posterior to the common genital pore, lead into the copulatory bursae, each having a narrow, long, bursal canal opening into the posterior part of the female genital canal separately from the ventral side; female genital canal long, wide, and opening into the common genital antrum anteriorly and into the cavity of the single seminal bursa posteriorly; seminal bursa connecting to an intestinal cross-link by a long, narrow, seminal bursa-intestinal duct;

ovovitelline ducts separately entering the middle part of the female genital canal (anterior part of the canal accompanied by erythrophilic shell glands); non-stalked cocoon spherical in shape (0.3–0.4 mm in diameter).

## ECOLOGICAL NOTES

Manoa Stream, the type locality of the present new species *Oahuhawaiiana kazukolinda*, is a narrow, stony stream and is about 3.5 km from Waikiki beach. A more detailed description of this locality and its photograph are found in a previous paper, together with the taxonomic description of *Dugesia dorotocephala* collected there [4].

According to Mr. Tanaka's observation, the amount of water in this stream varies considerably. Six cocoons were collected on 18 June 1966 at the type locality, and 4 juveniles were hatched from each of 2 of these within two weeks [1]. Although *D. dorotocephala* was also collected from two other localities in Honolulu, no specimens of the present new species were obtained from them [1, 2, 4].

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